

ORIGINAL ARTICLE

# Comparison of real-time PCR and cultural method for detection of bacterial load in pasteurized milk

Ayda Farhoudi<sup>1</sup> | Peyman Ghajarbeygi<sup>2</sup> | Razi Allah Jafari Jozani<sup>3</sup> |  
Razzagh Mahmoudi<sup>4</sup>  | Karim Mardani<sup>5</sup>

<sup>1</sup>School of Public Health, Qazvin University of Medical Sciences, Qazvin, Iran

<sup>2</sup>Health Products Safety Research Center, Qazvin University of Medical Sciences, Qazvin, Iran

<sup>3</sup>Department of Clinical Sciences, Faculty of Veterinary Medicine, University of Tabriz, Tabriz, Iran

<sup>4</sup>Medical Microbiology Research Center, Qazvin University of Medical Sciences, Qazvin, Iran

<sup>5</sup>Department of Food Hygiene and Quality Control, Faculty of Veterinary Medicine, Urmia University, Urmia, Iran

## Correspondence

Razzagh Mahmoudi, Qazvin University of Medical Sciences, Qazvin, Iran,  
Email: r.mahmoudi@yahoo.com

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Qazvin University of Medical Sciences

## Abstract

In this study, we used a TaqMan<sup>®</sup> probes through the real-time polymerase chain reaction (PCR) technique as a rapid and sensitive way to detect bacterial load of pasteurized milk. The reaction was optimized for enumeration of bacteria in pasteurized milk samples. In parallel, the same milk samples were assessed by conventional cultural-based method. The correlation between methods was evaluated by Bland Altman analysis. The minimum and maximum value for conventional culture-based method were 0 and 10,000 cfu/ml while qPCR gave us 55 and 7,071 (Bacteria/ml of pasteurized milk), respectively. Results indicated that Taq Man real-time PCR provides a useful tool for rapid and accurate quantification of bacterial load.

## Practical applications

Dairy industries are required to determine total bacterial load in farmer's raw milk before collection based on legal standard methods for microbial detection. Moreover, the result of total bacterial counting is the major pricing criterion of raw milk. Despite the storage of raw milk from large farms in separate tanks, raw milk from small- and medium-size farms are mixed in other tanks resulting in total bacterial load alteration. Traditional cultural-based methods are common to determine total bacterial load in raw milk. However, these techniques are laborious and time-consuming. Recent molecular-based methods are easily applicable to ensure that perishable raw milk could be monitored precisely under national rules and legislation in the shortest possible time. This report provides the details of a TaqMan real-time PCR (qPCR) that can be used in factory sites for accurate monitoring of raw milk bacterial load before transferring the raw milk into pasteurization line.

## 1 | INTRODUCTION

Milk is known as an excellent source of nutrient. It contains many valuable compounds such as vitamins and minerals and essential amino acids which have an important role in human health (Theresa, Nicklas, & Ph, 2003). Therefore, it is strictly recommended to every people whether young or old. As milk is a perishable product and might contain pathogens, it has the potential to cause foodborne diseases (Guh et al., 2010). Milk is a suitable medium to support the growth of some pathogenic bacteria such as *Escherichia coli* which is responsible for gastrointestinal infections in young children and elderly people (Blackall & Marques, 2004). So the proper quality

control of milk is important to make sure of its safety. Each country enforces legal standard methods for microbial detection in milk and in its products (Nanu, Latha, Sunil, et al., 2007). Conventional cultural-based methods are common to detect bacterial load in milk.

However, these methods are laborious and time-consuming, in most cases take up to almost 72 hr to cultivate (Gammon et al., 2007). Some dead and stressed cells and microorganism cannot be detected by the conventional method and some special incubation condition such as temperature and atmosphere may be needed for this method to be able to support the growth of those fastidious agents (Juste, Thomma, & Lievens, 2008). Food safety management system obligates that detection should get a result in the shortest possible time,